2018 NMR User Training Course (II)

How to process
2D & 3D NMR spectra
(Topspin 3.6)

By Alice Wei, Ph.D.

★ Videos for the process and plot of 1D and 2D spectra (Chinese version)
https://www.youtube.com/playlist?list=PLOFVd6zO7BsSAC7j0SKOAqEyQPFis_HEv
(google "youtube topspin 3.x", these videos are made by RA Huang, Yi-Ping)

2D Processing

@**The processing of 2D experiment (on TopSpin 3.6.0)---** Process

a) "**xfb**" (process data in F1 and F2 dimension), FID transfers to 2D spectrum.

b) Edit contour levels:

♦automatically (command mode)---

- Type "**clev 0.6 32**" to edit contour levels automatically (clev: automatically calculate levels for 2D data; 0.6: the factor for calculation; 32: number of levels).

♦manually---

- You can click the spectrum and scroll the mouse wheel (up or down) to adjust peaks' height you want to display.
- Type "**.lv**" or "**edlev**" (or click ) to contour levels mode, you can adjust "**level increment**" and "**number of levels**". After the adjustment, please click "**Fill**" and "**Apply**", and the contour values of positive and/or negative panels will be changed. At the same time, you can see the displayed cross-peaks will be changed in the spectrum. (Note: **normally, peaks will be positive in most spectra and you can ignore the Negative panel, but do not change the contour level sign to positive (keep in "Positive & Negative") before you finish the phase correction.**)

c) Phase correction:

♦Spectrum with window functions of "**QSINE**" in "**ProcPars**"---

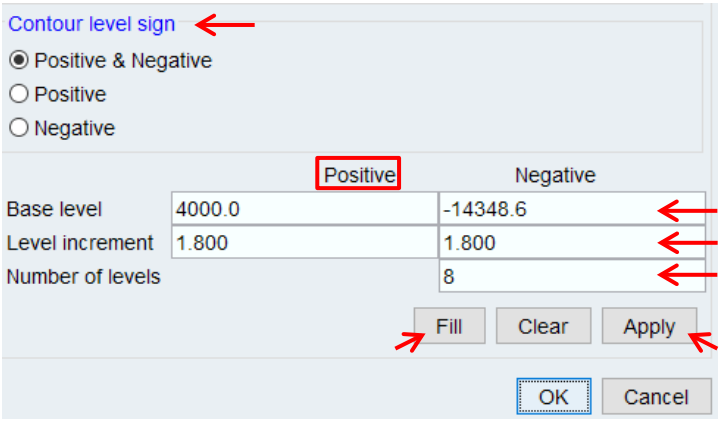
- Most of the spectra are with window functions of "**QSINE**". At first step, you check the value of "**2**" in **SSB** of "**ProcPars**" mode.
- Type "**.ph**" or click "**Adjust Phase**" to phase mode.
- In 2D spectrum, you need to select cross-peaks (normally 2~3 or 4, click the **right** mouse button to add/remove the peaks) in different area to do the phase correction in F2 axis (on row ) or F1 axis (on column ).
- Click "**0**" or "**1**" by using the **left** mouse button and drawing up or down to adjust the phase. These phase values will be recorded into "**ProcPars**" mode ("**edp**") after you finish the phase correction and save it.
  You can toggle grid mode () to make the grids in the background, and then use the grid to be a baseline's reference. Please click the spectrum, scroll the mouse wheel (up or down) to change the contour levels of cross-peaks, and then judge the phase correction.
- Change the value 2 to 2.5, 3, 3.5 or 4 in SSB of "**ProcPars**" mode.
- Click "**Positive**" of "**Contour level sign**" in contour levels mode for most spectra.

{Important: **Please view the PulseProg" to check if any particular requirement in the processing.**

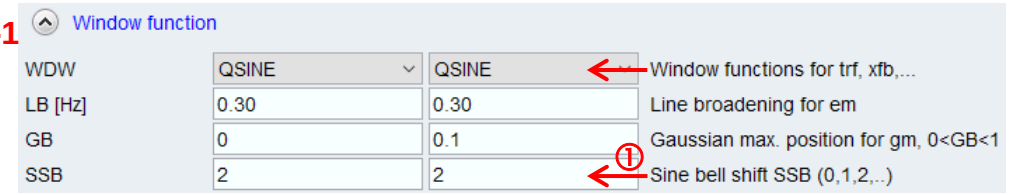
♦Spectrum with "**QF**" in F1 axis ("**FnMODE**") of "**AcquPars**"/"**PulseProg**"---

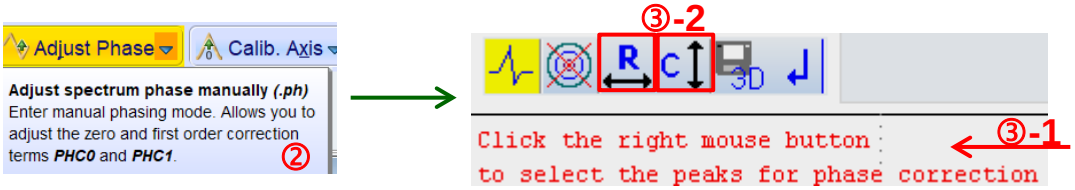
You don't need to do the phase correction. Only "**xfb**", edit contour levels, and baseline correction.

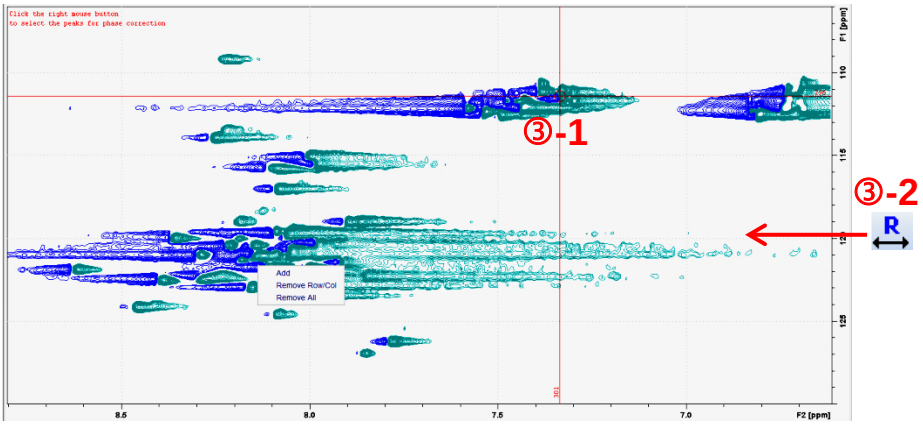
b)-2

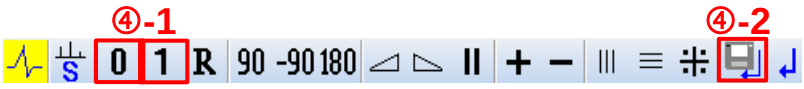

Contour level sign
Positive & Negative
Positive
Negative
Positive
Negative
Base level
4000.0
-14348.6
Level increment
1.800
1.800
Number of levels
8
Fill
Clear
Apply
OK
Cancel

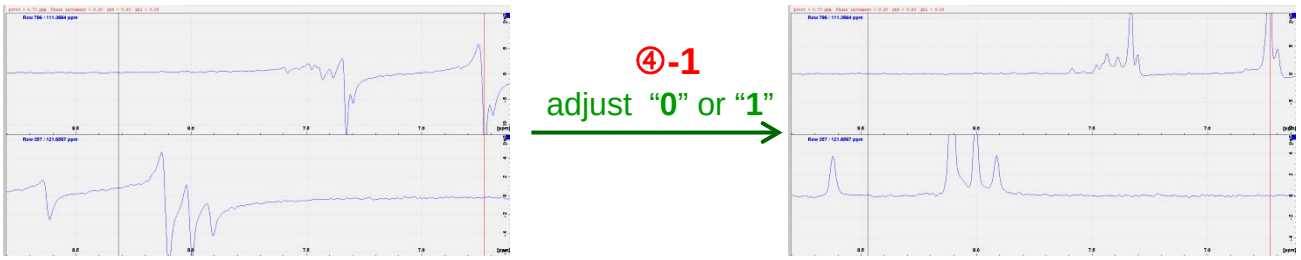
c)-1


Window function
WDW
QSINE
QSINE
Window functions for trf, xfb,...
LB [Hz]
0.30
0.30
Line broadening for em
GB
0
0.1
Gaussian max. position for gm, 0<GB<1
SSB
2
2
①
Sine bell shift SSB (0,1,2,..)

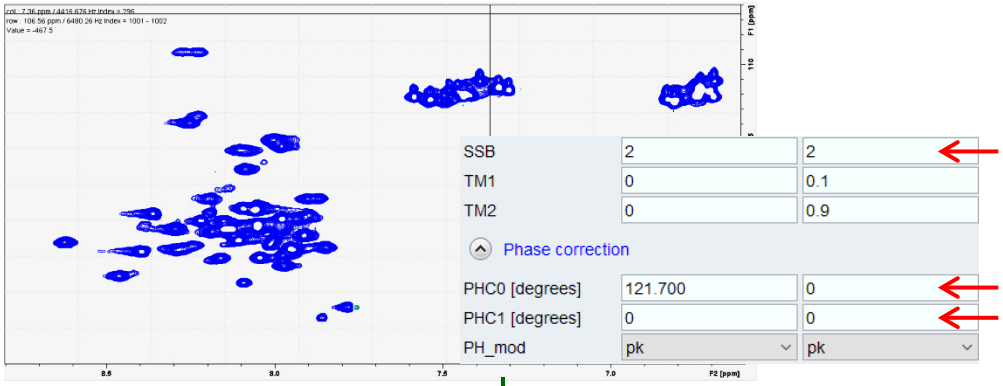

Adjust Phase
Calib. Axis
Adjust spectrum phase manually (.ph)
Enter manual phasing mode. Allows you to adjust the zero and first order correction terms PHC0 and PHC1.
②
③-2
R
C
3D
Click the right mouse button
to select the peaks for phase correction
③-1


③-1
③-2
R
Add
Remove Row/Col
Remove All
F2 [ppm]

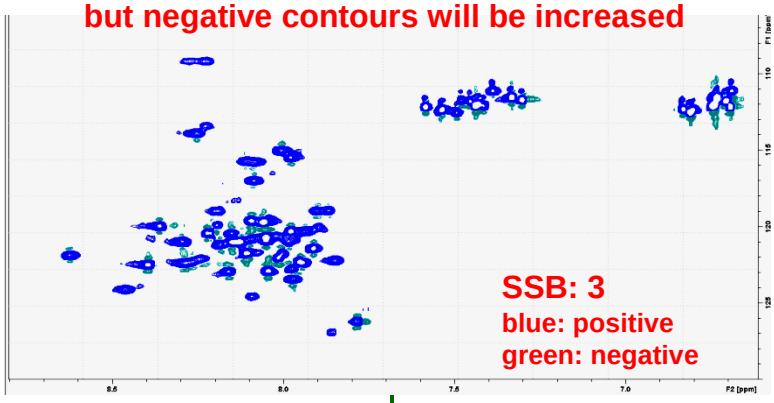

④-1
④-2
0
1
R
90
-90
180


④-1
adjust “0” or “1”

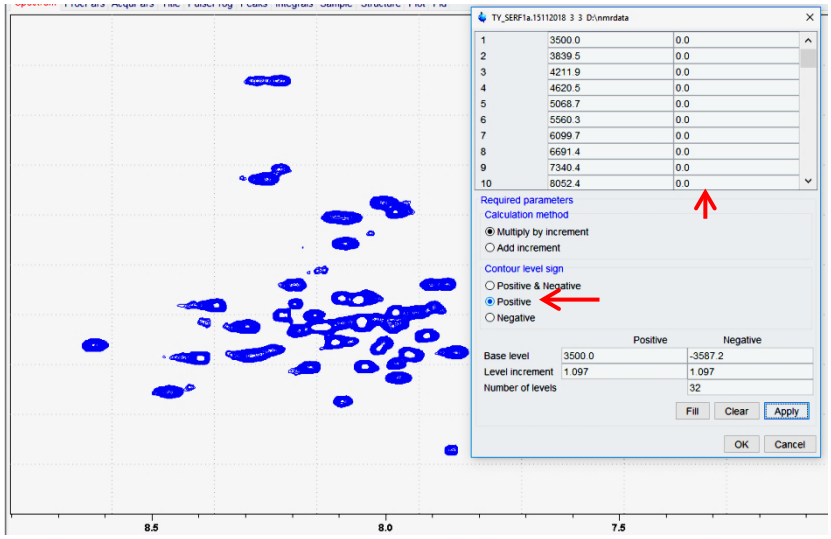
Repeat ③ and ④ steps until you get a good phase

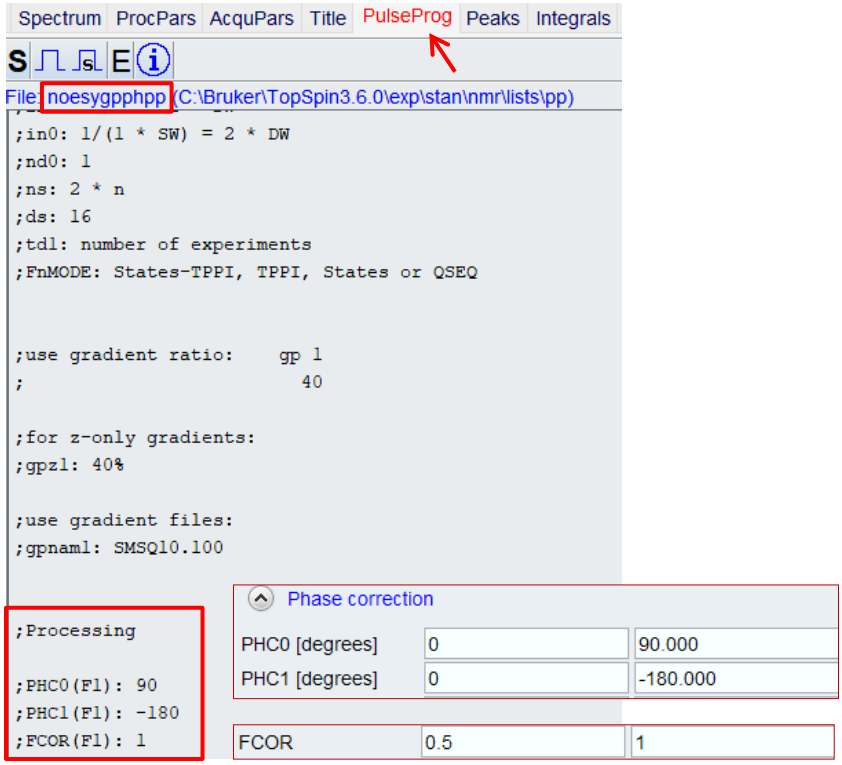


⑤Change the value 2 to 2.5, 3, 3.5 or 4 in SSB
=>The shape of cross-peaks are modified and the resolution is better, but negative contours will be increased

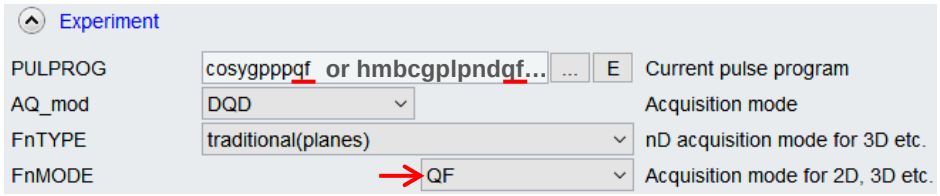


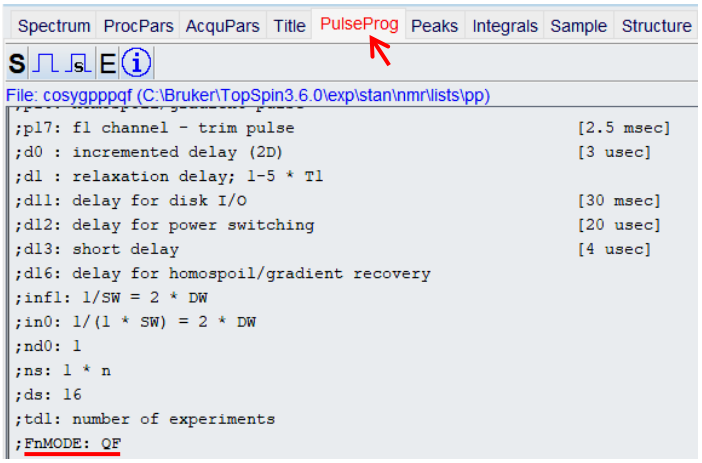
⑥Click “Positive” in “Contour level sign” to make all peaks positive for most spectra

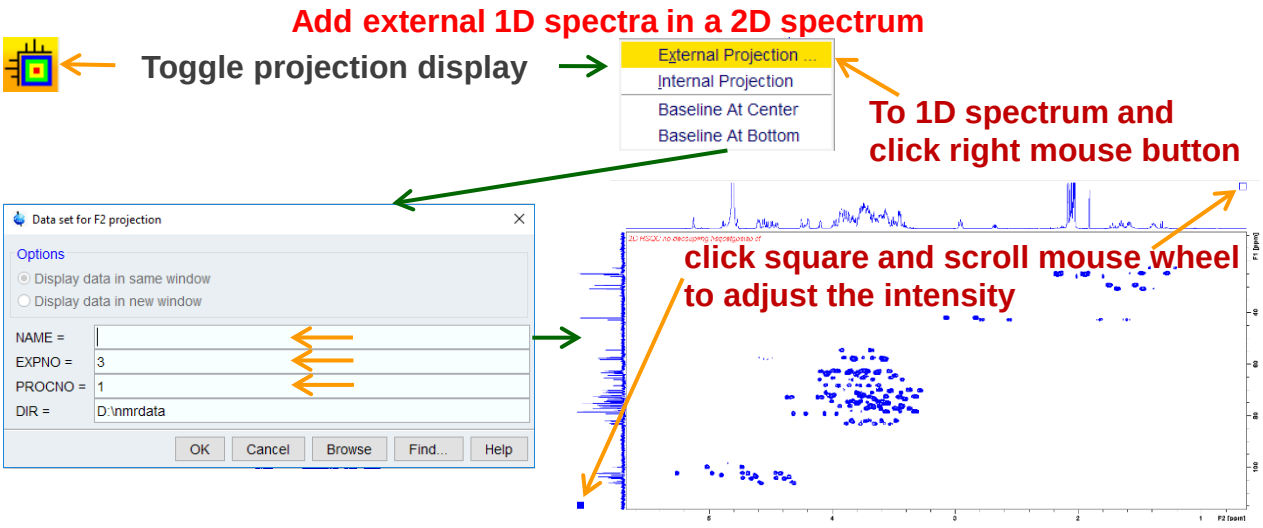



Spectrum ProcPars AcquPars Title PulseProg Peaks Integrals
File: noesygpphpp (C:\Bruker\TopSpin3.6.0\exp\stan\nmr\lists\pp)
;in0: 1/(1 * SW) = 2 * DW
;nd0: 1
;ns: 2 * n
;ds: 16
;tdl: number of experiments
;FnMODE: States-TPPI, TPPI, States or QSEQ
;use gradient ratio: gp 1
; 40
;for z-only gradients:
;gpz1: 40%
;use gradient files:
;gpnam1: SMSQ10.100
;Processing
;PHC0(F1): 90
;PHC1(F1): -180
;FCOR(F1): 1
Phase correction
PHC0 [degrees] 0 90.000
PHC1 [degrees] 0 -180.000
FCOR 0.5 1

c)-2


Experiment
PULPROG cosygpppqf or hmbcgplpndqf... E Current pulse program
AQ_mod DQD Acquisition mode
FnTYPE traditional(planes) nD acquisition mode for 3D etc.
FnMODE QF Acquisition mode for 2D, 3D etc.


Spectrum ProcPars AcquPars Title PulseProg Peaks Integrals Sample Structure
File: cosygpppqf (C:\Bruker\TopSpin3.6.0\exp\stan\nmr\lists\pp)
;p17: f1 channel - trim pulse [2.5 msec]
;d0 : incremented delay (2D) [3 usec]
;d1 : relaxation delay; 1-5 * T1
;d11: delay for disk I/O [30 msec]
;d12: delay for power switching [20 usec]
;d13: short delay [4 usec]
;d16: delay for homospoil/gradient recovery
;inf1: 1/SW = 2 * DW
;in0: 1/(1 * SW) = 2 * DW
;nd0: 1
;ns: 1 * n
;ds: 16
;tdl: number of experiments
;FnMODE: QF



d) Calibration: Click "**ProcPars**" or type "**edp**" and key in the calibration values in F1 and F2 of SR [Hz] (spectrum reference frequency. (Note: **please pay attention on nucleus type when you get the SR value from 1D experiments or guideline handbook**).

e) Baseline correction:

- In "**ProcPars**" mode ("**edp**"), you can key the values (in ppm) into ABSF1 and ABSF2 in F1 and F2 axes to set the range you want to do the baseline correction.
- Type "**abs1**" (automatic baseline correction in F1 axis).
- Type "**abs2**" (automatic baseline correction in F2 axis).

d)

Reference	F2	F1	
SI	2048	1024	Size of real spectrum
SF [MHz]	600.1300000 ^{1}H	60.8106450 ^{15}N	Spectrometer frequency
OFFSET [ppm]	8.80300	130.00140	Low field limit of spectrum
SR [Hz]	0	0	Spectrum reference frequency

e)

Baseline correction	F2	F1	
ABSG	5	5	Degree of polynomial for abs (0..5)
ABSF1 [ppm]	100.00000	1000.00000	Left limit for absf
ABSF2 [ppm]	-100.00000	-1000.00000	Right limit for absf, abs1, abs2
BCFW [ppm]	1.00000	1.00000	Filter width for bc (sfil/qfil)
COROFFS [Hz]	0	0	Correction offset for BC_MOD=spol etc.
BC_mod	qpol	no	Fid baseline modes for em, ft, xfb,...

★ **Videos for the process and plot of 1D and 2D spectra (Chinese version)**
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3D Processing

@The processing of 3D experiment (on TopSpin 3.6.0)--- Process

a) Click "**ProcPars**" (or type "**edp**") to process parameters' window and set the parameters (SI, PHC0, PHC1, ABSF1, ABSF2, etc. in F1, F2 and F3 axes)

b) "**xfb**" to process data → Select plane axis orientation (**23** or **13**) → Enter plane number (normally, **1**) → Enter new PROCNO (process number) for 2D data

c) Edit contour levels. Please refer to **the processing of 2D**.

d) Phase correction. Please refer to **the processing of 2D**. After you finish the phase correction in F2F3 (23) plane and F1F3 (13) plane, please copy the values of PHC0 and PHC1 in F1, F2 and F3 axes to the 3D experiment. (Note: **Only one F3 axis in a 3D experiment. PHC0 and PHC1 of F3 should be the same in F2F3 and F1F3 planes.**)

e) Click "**ProcPars**" (or type "**edp**") and set or check the parameters (SR, PHC0, PHC1, ABSF1, ABSF2, STSR, STSI, etc. in F1, F2 and F3 axes) in a 3D spectrum.

f) "**ft3d n**" to process 3D data, including FT, without imaginary file (only 3rrr file in pdata folder).

g) Type "**tabs3**", "**tabs2**" and "**tabs1**" to do the baseline correction, repectively.

SI ≥ 2X TD

	F3	F2	F1	
SI	2048	256	512	Size of real spectrum
SF [MHz]	800.2400000	81.0876150	201.2205080	Spectrometer frequency
OFFSET [ppm]	11.67507	133.00180	73.00131	Low field limit of spectrum
SR [Hz]	0	0	0	Spectrum reference frequency
HZpPT [Hz]	2.554485	9.503576	25.153595	Spectral resolution
AQORDER	3-2-1			Order of fids in ser file
SPECTYP	UNDEFINED			Type of spectrum e.g. COSY, HMQC, ...
Window function				
WDW	QSINE	QSINE	QSINE	Window functions for trf, xfb,...
LB [Hz]	1.00	0.30	0	Line broadening for em
GB	0.1	0.1	0	Gaussian max. position for gm, 0<GB<1
SSB	2	3	3	Sine bell shift SSB (0,1,2,..)
TM1	0.1	0.1	0.1	Left limit for tm 0<TM1<1
TM2	0.9	0.9	0.9	Right limit for tm 0<TM2<1
Phase correction				
PHC0 [degrees]	-78.000	0	0	0th order correction for pk
PHC1 [degrees]	0	0	0	1st order correction for pk
PH_mod	pk	pk	pk	Phasing modes for trf, xfb, ...

PHC0/PHC1 should be zero and SSB = 2 before doing the phase correction of 2D spectrum.

Pulse program

```
;Processing

;SR(F1): 1/4 SWH(F1)
```

SR(F1) = SR_H + ¼ SWH (F1)
in HCCH-TOCSY, HBHA(CO)NH, etc.

Baseline correction

	F3	F2	F1	
ABSG	5	5	5	Degree of polynomial for abs (0..5)
ABSF1 [ppm]	100.00000	1000.00000	1000.00000	Left limit for absf
ABSF2 [ppm]	-100.00000	-1000.00000	-1000.00000	Right limit for absf, abs1, abs2

Fourier transform

TDeff	0	0	0	Number of fid data points used by ft
STSR	0	0	0	First output point of strip transform
STSI	950	0	0	Total number of output points of strip transform
ME_mod	no	LPfc	no	Linear prediction for ft, xfb, ...
NCOEF	0	32	0	Number of LP coefficients
LPBIN	0	0	0	Number of output points for LP
TDoff	0	0	0	Number of back-predicted points
REVERSE	FALSE	FALSE	TRUE	Reverse spectrum during transform
FCOR	0.5	0.5	0.5	Weighting factor for first fid point

>TD_{eff} = TD or 0

>Set STSR/STSI to remove the blank space (no data region)

>linear prediction in the axis with poor FID resolution. {ME_mod: LPfc (most used); NCOEF: 8 * n}